

EVALUATION OF METHODS OF REDUCING SEED DORMANCY
IN INDIANGRASS, SORGHastrum nutans (L.) NASH

by

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INTRODUCTION

Indiangrass, Sorghastrum nutans (L.) Nash, is a tall, coarse, erect, deep-rooted, native perennial. It has been reported to produce excellent feed either as hay or pasture (Wheeler and Hill, 1957). This grass has a wide range throughout the eastern and central United States and in parts of Canada and Mexico (Anonymous, 1954).

Establishment and seed testing of this grass are complicated by high levels of seed dormancy which frequently persist a year or more after seed harvest. The primary objective of this study was to evaluate the dormancy-breaking effects of various germination and pregermination treatments upon seed of indiangrass. It was hoped that results of the study would also help to elucidate the mechanism of seed dormancy of this species.

The nature and probable causes of seed dormancy have been discussed by Hartmann and Kester (1959); Meyer, Anderson, and Böhning (1960); Toole (1961); and Pollock and Toole (1961). In this study seed dormancy will be interpreted as germination failure in viable seeds resulting from conditions within the caryopsis or the chaffy appendages normally adhering to the caryopsis.

LITERATURE REVIEW

Treatments for Overcoming Seed Dormancy

Soaking in Water

The leaching effect of water has been found effective in reducing seed dormancy in some species. Rogler (1960 a, 1960 b) reported that seeds of green needlegrass, Stipa viridula Trin., and indian ricegrass; Oryzopsis hymenoides (Roem. and Schult) Ricker., showed a marked increase in germination when soaked in water at 2-4°C for 20 and 40 days respectively. Sumner and Cobb (1962) found that a 30-minute washing of seeds of Coronado side-oats grama, Bouteloua curtipendula (Mich.) Torr., increased germination from 64% to 83%. Elliot and Leopold (1953) reported that washing with water led to stimulation of germination in seeds of Victory oats and that there was a concomitant appearance of an inhibitor of α -amylase in the eluted solutions.

Soaking in Aqueous Solution of Hydrogen Peroxide

Demowsay (1916) reported a significant increase in germination of seed of garden cress, Lepidium sativum L., following soaking in hydrogen peroxide solution.

Treatment with Gibberellic Acid

Gibberellic acid treatments have been shown to overcome some types of dormancy. Presoaking in aqueous gibberellin solution (10-250 ppm) was reported by Merck and Co., Inc. (1957) to accelerate germination of seeds of oats, maize, and other species.

Button (1959) found that gibberellic acid treatment was effect in hastening seed germination of creeping red fescue, Festuca rubra L., during the early part of the standard test period. Seeds of ryegrass, tall fescue, and Kentucky bluegrass have shown similar response to gibberellic acid. According to Naylor and Simpson (1961), natural inhibition of germination in wild oats, Avena fatua L., has been overcome by treatment with gibberellic acid.

Soaking in Aqueous Solution of Sodium Hypochlorite (Clorox)

Sodium hypochlorite (Clorox) has been reported effective in destroying germination inhibitors. Went (1957) reported that this compound could destroy the seedcoat inhibitor and improve seed germination of Pectis papposa Harv. and Gray. Summner and Cobb (1962) found that treatment of seed units of Coronado side-oats grama with Clorox overcame the effects of inhibiting substances hindering germination but neither impaired nor improved germination potential.

Exposure to Low Temperature

Low temperature treatment has been recognized as an effective means of reducing seed dormancy in many species. Blake (1935) reported that stratification of seeds of indian-grass through the winter months resulted in a marked increase in germination. Barton (1939) reported that, with some species, stratification was one of the best methods for inducing germination of seeds with dormant embryos. Kearns and Toolc (1939) found

that good germination of fescue seed followed prechilling for seven days at 5°C. According to Crocker (1948), low temperature treatment of imbibed seeds has commonly been used to induce after-ripening. Freshly harvested dormant seeds of cereals have been induced to germinate by a few days of prechilling, or by subjection to low temperature during germination. Other dormant grass seeds have been found to respond to stratification or to prechilling. Prechilling at 5°C for two weeks is part of the standard treatment for breaking seed dormancy of indiangrass (Assoc. Off. Seed Anal. 1960).

Exposure to High Temperature

In most cases, high temperature has been found to increase seed dormancy rather than reduce it (Leopold, 1964). However, Ahring, Dunn, and Harlan (1963) found that preheating seed of sand lovegrass, Eragrostis trichodes (Nutt.) Wood, for 30 or 40 minutes at 90° to 100°C was effective in breaking dormancy. Preheating for periods of 12 and 24 hours at 50° to 60°C was also effective. Mayer and Poljakoff-Mayber (1963) suggested that high temperature could cause a change in seed coat permeability.

Hull Removal

Removal of the chaffy coverings of the caryopsis has been shown effective in reducing seed dormancy in a number of grasses. Ray and Stewart (1937) and Barton (1939) found hulling effective in increasing germination of several species of Paspalum. Forbes and Ferguson (1948) obtained similar results with Zoysia japonica.

Steud. Dawson and Heinricks (1952) reported increased seed germination following complete removal of the lemma and palea of green stipagrass, Stipa viridula Trin. They suggested that the lemma and palea were a barrier to water uptake. Fendall and Carter (1965), however, reported that hull removal resulted in an immediate acceleration of oxygen utilization in relatively dormant seed of green stipagrass. Roberts (1961) found partial or complete removal of hulls effective in improving germination of rice. Canode, Horning, and Maguire (1963) reported that removal of the lemma and palea in the threshing process gave a significant increase in germination of seed of orchardgrass, Dactylis glomerata L.

MATERIALS AND METHODS

Seed Lots

Lot A

This seed was harvested in 1960 from the first synthetic generation of Kansas Agricultural Experiment Station strain 1. Strain 1 is a synthetic of 45 clones which originated as seed in parts of Kansas, Oklahoma and Texas. Harvest was by combining and the seed was subsequently cleaned through scalping, fanning, and screening.

Lot B

This seed was harvested in 1963 from the first synthetic generation of Kansas Agricultural Experiment Station strain 2. Strain 2 is a synthetic of 20 clones selected from those of strain 1. Harvest and cleaning were as described for Lot A.

Lot C

Seed of this lot was harvested in 1962 from the first synthetic generation of Kansas Agricultural Experiment Station strain 1. The seed was combined and cleaned in the manner described for Lot A.

Lot D

This lot was harvested in 1963 from the first synthetic generation of Kansas Agricultural Experiment Station strain 3. Strain 3 is a synthetic of eight clones selected from those of strain 2. The seed was combined and cleaned in the manner

described for Lot A.

Lot E

This seed was harvested in 1963 from a foundation field of the variety, Cheyenne. Cheyenne was developed from materials collected from native rangelands near Supply, Oklahoma. The seed was combined but only partially cleaned.

Lot F

This seed was harvested in 1964 from the first synthetic generation of Kansas Agricultural Experiment Station strain 4. Strain 4 is a synthetic of six clones selected from those of strain 2. The seed was combined but not cleaned.

Lot G

This lot was harvested in 1965 from the zero synthetic generation of strain 3. Harvest was by hand, and the harvested material was not cleaned.

Lot H

This lot was harvested in 1965 from the first synthetic generation of strain 3. The seed was combined and cleaned in the manner described for lot A.

All harvests were made in the vicinity of Manhattan, Kansas. After being obtained, all seed lots were stored at room temperature under dry conditions.

Sampling

The indiangrass spikelet possesses one perfect and one staminate floret and is normally capable of producing only one caryopsis. All germination tests involved samples of 100 randomly selected spikelets.

Pregermination Treatments

Exposure to Low Temperature and Room Temperature for Fourteen Days under Varying Moisture Conditions

High Dry. Samples of 100 randomly selected spikelets were maintained in paper envelopes at room temperature and under dry conditions. The high dry pretreatment constituted the no-treatment check in this and subsequent tests.

High Immersion. Samples were immersed in approximately 20 ml of tap water in 25-ml vials and maintained at room temperature. The water was changed every three days.

Low Dry. Samples were maintained in paper envelopes at approximately 5°C.

Low Moist. Samples were placed on two layers of moistened filter paper 9-cm plastic petri dishes and maintained in a moistened condition at approximately 5°C.

Low Immersion. Samples were immersed in approximately 20 ml of tap water in 25-ml glass vials and kept at approximately 5°C. The water was changed every three days.

Seed lots A, B, C, D, E, and F were subjected to the above treatments. Duration of all treatments was 14 days immediately prior to the germination test. This experiment was conducted in April, 1965.

Exposure to Low Temperature and Room Temperature for Varying Periods under Varying Moisture Conditions

Samples of lots D and E were subjected to the high immersion, low moist and low immersion pretreatments (as described above) for 1, 3, 7 and 14 days. Additional samples were subjected to the high dry pretreatment for 14 days prior to the germination test. This experiment was conducted in May, 1965.

Exposure to High Temperature

Dry samples of lots E and H were placed in an oven at 50°-60°C for 15, 30, 45, 60 and 75 minutes. Other samples were maintained at 90°-100°C for the same periods. Additional samples were subjected to the high dry and low moist pretreatments. This experiment was conducted in March, 1966.

Soaking in Aqueous Solution of Hydrogen Peroxide

Samples of lots E and H were soaked in a 1.0% solution of hydrogen peroxide for 1, 2, 4, 8, 15, 30, and 60 minutes and in a 5.0% solution of hydrogen peroxide for 15, 30, and 60 minutes. Additional samples were soaked in tap water for 60 minutes while others were subjected to the high dry and low moist pretreatments (as previously described) for 14 days. This experiment was conducted from February through March in 1966.

Soaking in Aqueous Solution of Sodium Hypochlorite (Clorox)

Clorox, a commercial product containing 5.25% sodium hypochlorite (NaOCl), was used in this test.

Samples of lots E and H were soaked in Clorox for 15, 30, 45, and 60 minutes. Additional samples were soaked in distilled water for 60 minutes. Other samples were subjected to the high dry and low moist pretreatments for 14 days. Spikelets treated with Clorox were rinsed three times in tap water prior to the germination test. This experiment was conducted from April through May in 1966.

Hull Removal

Hulls of randomly selected spikelets from lots E and G were removed through use of a corrugated rubbing board. Additional samples (entire spikelets) were subjected to the high dry and low moist pretreatments for 14 days. This experiment was conducted from January through February in 1966.

Germination Test

Following the pregermination treatments, each sample was placed in a plastic petri dish on two layers of filter paper and moistened with approximately 5 ml of tap water. The dishes were then placed in a germinator at approximately 25°C.

Spikelets were considered to have germinated normally when both plumule and radicle had reached a length of approximately 5 mm. Germination counts were made every two days for approximately three weeks. Germinated spikelets were removed from the petri

dishes when counted. Spikelets were maintained in a moist condition throughout the germination test.

Treatment with Gibberellic Acid

Samples of lots E and H were placed on two layers of filter paper in petri dishes and moistened with 5 ml of aqueous solution of gibberellic acid. Concentrations of gibberellic acid of 25, 50, 75 and 100 mg/liter were used. The petri dishes were placed in a germinator at approximately 25°C, and subsequent procedures were the same as those in the regular germination test. Samples subjected to the high dry and low moist pregermination treatments were used as checks. This experiment was conducted from March through April in 1966.

Relation of Caryopsis Moisture Uptake to Seed Dormancy

Samples of free caryopses of lots E and H were obtained through hand removal of the chaffy appendages of spikelets. Samples of 100 caryopses were weighed, soaked in tap water for 24 hours, and weighed again. They were then placed in an oven at 90°-100°C and weighed to constant weight. The initial weight, the weight after soaking, and the weight after oven drying were recorded. This experiment was conducted in May, 1966.

Statistical Design and Analysis

All germination tests were designed as randomized complete block experiments with four replications, each replication consisting of a separate tray in the germinator. Germination percentages were transformed to $\text{arc sin } \sqrt{\text{percentage}}$ (Snedecor, 1956) before analysis.

Data on caryopsis moisture uptake, in relation to seed dormancy, were analyzed according to the following formula (Li, 1957).

$$u = \frac{\bar{y}_1 - \bar{y}_2}{\sqrt{\frac{\bar{y}(1-\bar{y})}{n_1 + n_2} \left(\frac{1}{n_1} + \frac{1}{n_2} \right)}}$$

where

\bar{y}_1 = Mean moisture uptake (% dry weight expressed as a decimal fraction) of first seed lot.

\bar{y}_2 = Mean moisture uptake (% dry weight expressed as a decimal fraction) of second seed lot.

$\bar{\bar{y}}$ = Arithmetic mean of the two sample means.

n_1 = Number of replications of first seed lot.

n_2 = Number of replications of second seed lot.

A value of u greater than 1.96 indicated significance at the 0.05 level. Percent moisture uptake was based on both initial weight and the weight following oven drying. The number of replications of each seed lot was four.

RESULTS

Exposure to Low Temperature and Room Temperature for Fourteen Days under Varying Moisture Conditions

Results are summarized in Tables 1-3. Data for seed lot F were not included in the analysis of variance as germination of this lot was very low and included many zero values (Table 3). Highly significant differences occurred among seed lots and among pretreatments. The seed-lot-by-pretreatment interaction was also highly significant. In mean germination for all seed lots only the low moist pretreatment resulted in a significant improvement over the high dry check. Improvement following the low immersion pretreatment approached significance at the 0.05 level.

With lot A, the low dry pretreatment yielded the highest germination, but germination following this pretreatment was not significantly superior to that of the high dry check. The high immersion pretreatment resulted in a significant reduction in germination. In lots B and D, the low moist and low immersion pretreatments yielded significant increases in germination while in lots C and E no pretreatment was significantly superior to the high dry check. The low moist pretreatment appeared to give the best results in lot F. The high dry and low dry pretreatments did not differ significantly in any seed lot.

Table 1. Analysis of variance of germination following exposure to low temperature and room temperature for 14 days under varying moisture conditions (data from lot F excluded).

| Source | D.F. | S.S. | M.S. | F. |
|----------------------|------|----------|---------|----------|
| Total | 99 | 29470.61 | 297.69 | |
| Treatment | 24 | 27596.09 | 1149.84 | 45.70** |
| Lots | (4) | 25071.66 | 6267.92 | 245.12** |
| Pretreatments | (4) | 969.78 | 242.45 | 9.64** |
| Lots X Pretreatments | (16) | 1554.65 | 97.17 | 3.86** |
| Blocks | 3 | 62.80 | 20.93 | 0.83 |
| Error | 72 | 1811.72 | 25.16 | |

* Significant at 0.05 level.

** Significant at 0.01 level.

Table 2. Mean germination (arc sin $\sqrt{\%}$) following exposure to low temperature and room temperature for 14 days under varying moisture conditions.

| Pretreatment | Within seed lots | | | Over all seed lots | | |
|----------------|------------------|------|------|--------------------|------|------|
| | A | B | C | | | |
| High dry | 17.7 | 41.8 | 20.1 | 32.9 | 54.9 | 33.5 |
| High immersion | 10.0 | 44.1 | 19.0 | 36.6 | 51.1 | 32.5 |
| Low dry | 21.4 | 42.4 | 18.2 | 32.2 | 54.8 | 33.8 |
| Low moist | 13.5 | 54.0 | 21.5 | 53.2 | 59.2 | 40.3 |
| Low immersion | 15.9 | 55.0 | 17.4 | 45.2 | 59.7 | 38.3 |

L.S.D. for comparing pretreatment means within seed lots,

at 0.05 level = 7.1.

at 0.01 level = 9.4.

L.S.D. for comparing pretreatment means over all seed lots,

at 0.05 level = 5.0.

at 0.01 level = 6.7.

C.V. = 48.5%.

Table 3. Germination of seed lot F.

| Pretreatment | Mean arc sin $\sqrt{\%}$ |
|----------------|-----------------------------|
| High dry | 3.14 |
| High immersion | 5.13 |
| Low dry | 5.14 |
| Low moist | 12.25 |
| Low immersion | 4.05 |

Exposure to Low Temperature and Room
Temperature for Varying Periods under
Varying Moisture Conditions

Results are summarized in Tables 4 and 5 and in Figures 1 and 2. In the analysis of variance, durations of pretreatments are referred to as times, while the various combinations of temperature and moisture availability are referred to as, conditions. The high dry pretreatment is the check.

Highly significant differences occurred between seed lots, among times, and among conditions. The interactions: times by conditions, seed lots by conditions, and seed lots by times by conditions were also highly significant.

The high immersion pretreatment did not significantly increase germination in either seed lot, although germination increase following pretreatment for one day approached significance at the 0.05 level in lot D. High immersion pretreatment for long periods reduced germination in both seed lots.

Low moist pretreatment for three days or longer significantly improved germination in lot D but not in lot E. Similar results followed the low immersion pretreatment, although here germination increase following pretreatment for one day approached significance at the 0.05 level in lot D. Where longer durations of pretreatment were involved, the low moist pretreatment seemed somewhat more effective than the low immersion pretreatment.

Table 4. Analysis of variance of germination following exposure to room temperature and low temperature for varying periods under varying moisture conditions.

| Source | D.F. | S.S. | M.S. | F |
|--------------------------------|------|---------|---------|----------|
| Total | 103 | 6217.36 | 60.36 | --- |
| Treatments | 25 | 5295.32 | 211.81 | 17.35** |
| Seed Lots | 1 | 1834.56 | 1834.56 | 150.25** |
| Pretreatments | 12 | 2940.80 | 245.07 | 20.07** |
| Times | (3) | 112.41 | 37.47 | 3.07** |
| Conditions | (2) | 1867.65 | 933.82 | 76.48** |
| Check vs. Others | (1) | 14.37 | 14.37 | 1.18 |
| Times X Conditions | (6) | 946.37 | 157.73 | 12.92** |
| Seed Lots X Pretreatments | 12 | 519.96 | 43.33 | 3.55** |
| Seed Lots X Times | (3) | 56.25 | 18.75 | 1.54 |
| Seed Lots X Conditions | (2) | 192.64 | 96.32 | 7.89** |
| Seed Lots X (Check vs. Others) | (1) | 35.09 | 35.09 | 2.87 |
| Seed Lots X Times X Conditions | (6) | 235.98 | 39.33 | 3.22** |
| Blocks | 3 | 6.14 | 2.05 | 0.17 |
| Error | 75 | 915.90 | 12.21 | |

* Significant at 0.05 level.

** Significant at 0.01 level.

Table 5. Mean germination (arc sin $\sqrt{\%}$) following exposure to room temperature and low temperature for varying periods under varying moisture conditions.

| Pretreatment | Lot D | Lot E | Over both seed lots |
|----------------|-------|-------|---------------------|
| High dry | 44.4 | 56.8 | 50.6 |
| High immersion | | | |
| 1 day | 49.8 | 56.7 | 53.3 |
| 3 days | 44.0 | 53.6 | 48.8 |
| 7 days | 32.3 | 53.6 | 43.0 |
| 14 days | 34.4 | 44.2 | 39.3 |
| Low moist | | | |
| 1 day | 47.9 | 61.6 | 54.8 |
| 3 days | 54.6 | 59.7 | 57.2 |
| 7 days | 54.6 | 58.5 | 56.6 |
| 14 days | 56.4 | 58.5 | 57.5 |
| Low immersion | | | |
| 1 day | 49.2 | 55.7 | 52.5 |
| 3 days | 51.3 | 57.4 | 54.4 |
| 7 days | 49.9 | 58.5 | 54.2 |
| 14 days | 51.4 | 58.0 | 54.7 |

L.S.D. for comparing pretreatment means within seed lots,
at 0.05 level = 5.5.
at 0.01 level = 6.5.

L.S.D. for comparing pretreatment means over both seed lots,
at 0.05 level = 5.9.
at 0.01 level = 4.6.

C.V. = 14.9%.

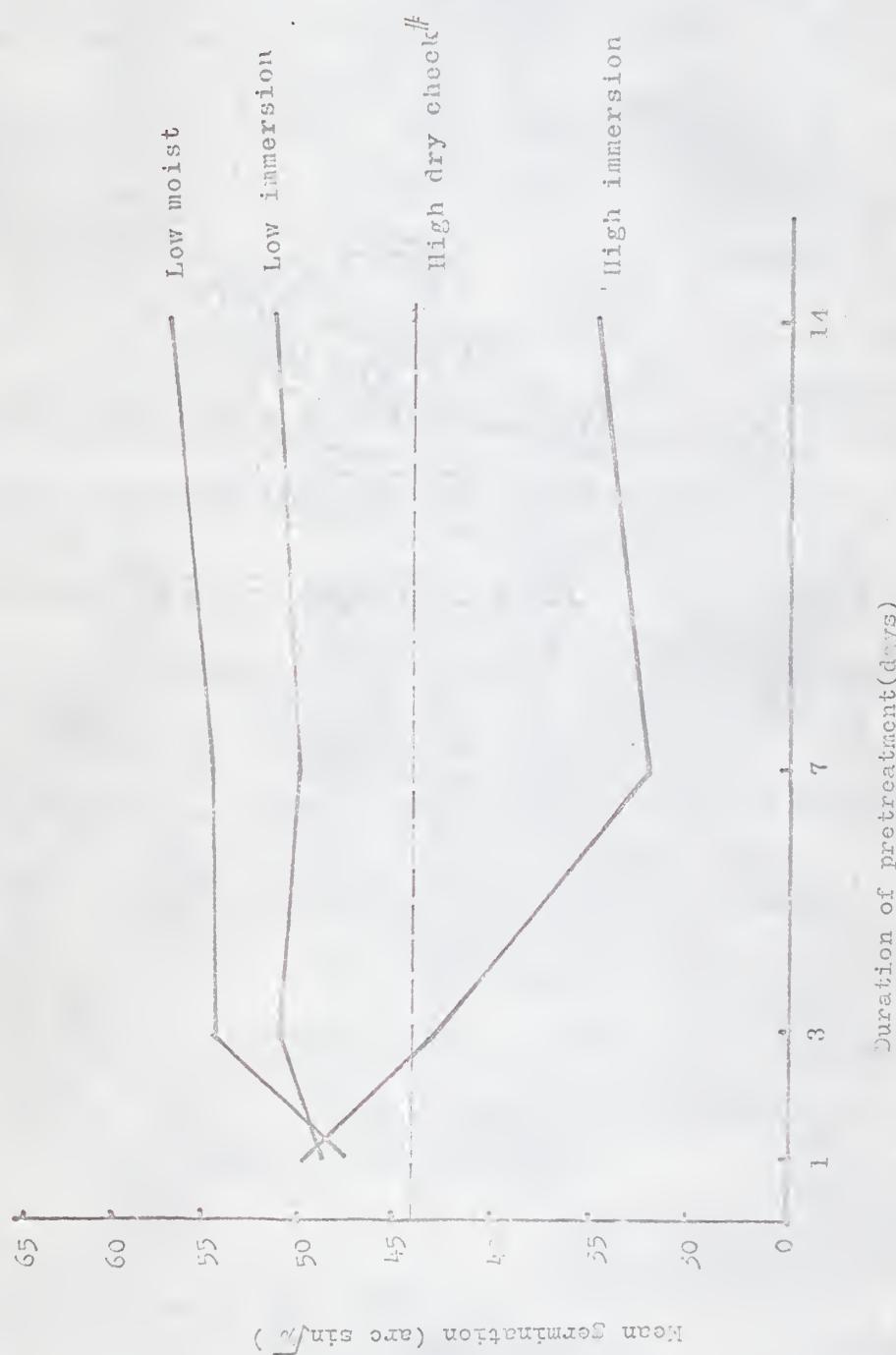


Fig. 1. Mean germination following exposure to room temperature and low temperature for varying periods under varying moisture conditions in seed lot D.

for 14 days.

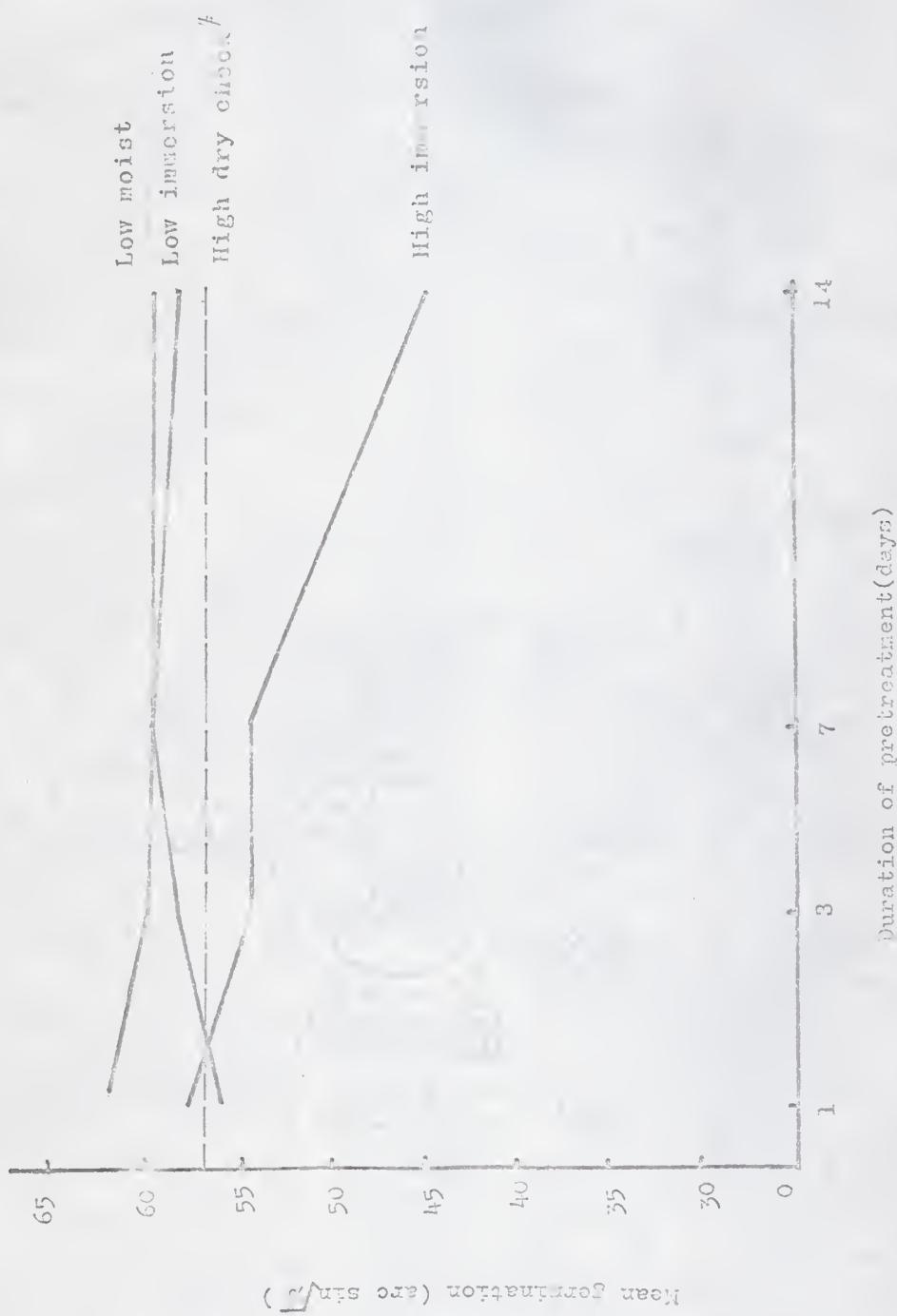


Fig. 2. Mean germination following exposure to room temperature and low temperature for varying periods under varying moisture conditions in seed lot 5, for 14 days.

Exposure to High Temperatures

Results are summarized in Tables 6 and 7. In the analysis of variance durations of exposure are referred to as times while the high dry and low moist pretreatments are referred to as checks.

Exposure to high temperature did not significantly improve germination in either seed lot. Exposure to temperatures of 90-100°C for 30, 45, 75, and 90 minutes resulted in a significant reduction in germination in lot E but not in lot H.

The low moist pretreatment significantly improved germination in lot H but not in lot E.

Soaking in Aqueous Solution of Hydrogen Peroxide

Results are summarized in Tables 8 and 9. Highly significant differences occurred between seed lots and among pretreatments. The lot-by-pretreatment interaction was also highly significant.

Soaking in aqueous solution of hydrogen peroxide yielded erratic results. Significant germination increases were obtained in a few instances with lot E, but no definite trend with respect to concentration or duration of pretreatment was evident. Germination of lot H was not significantly affected by the hydrogen peroxide pretreatment.

The low moist pretreatment significantly improved germination in both seed lots, while soaking in water made no improvement in either. In no case was germination following the hydrogen

Table 6. Analysis of variance of germination following exposure to high dry and low moist pretreatments.

| Source | D.F. | S.S. | M.S. | F |
|----------------------------------|------|----------|----------|-----------|
| Total | 111 | 44584.63 | 401.66 | |
| Treatments | 27 | 43547.94 | 1612.87 | 138.21** |
| Seed Lots | 1 | 42669.94 | 42669.94 | 3656.38** |
| Pretreatments | 13 | 509.72 | 39.21 | 3.36** |
| Between Checks | (1) | 43.56 | 43.56 | 5.73 |
| Between Temperatures | (1) | 159.39 | 159.39 | 13.66** |
| Among Times | (5) | 40.00 | 8.00 | 0.69 |
| Temperature X Times | (5) | 61.17 | 12.32 | 1.05 |
| Check vs. Others | (1) | 205.60 | 205.60 | 17.62** |
| Seed Lots X Pretreatments | 13 | 368.31 | 28.33 | 2.43** |
| Seed Lots X Checks | (1) | 85.57 | 85.57 | 7.33** |
| Seed Lots X Temperatures | (1) | 106.05 | 106.05 | 9.09** |
| Seed Lots X Times | (5) | 108.52 | 21.70 | 1.86 |
| Seed Lots X Check vs. Others | (1) | 4.32 | 4.32 | 0.37 |
| Seed Lots X Temperatures X Times | (5) | 63.85 | 12.77 | 1.09 |
| Blocks | 3 | 91.15 | 30.38 | 2.60 |
| Error | 81 | 945.51 | 11.67 | |

* Significant at 0.05 level.

** Significant at 0.01 level.

Table 7. Mean germination (arc sin $\sqrt{\%}$) following exposure to high temperature, and the high dry and low moist pretreatments.

| Pretreatment | Within seed lots | | Over both seed lots |
|--------------------------------|------------------|-------|---------------------|
| | Lot E | Lot H | |
| High dry | 58.2 | 15.5 | 36.9 |
| Low moist | 56.9 | 23.4 | 40.2 |
| 50° - 60° C | | | |
| 15 min. | 56.3 | 15.7 | 36.0 |
| 30 min. | 59.3 | 16.4 | 37.9 |
| 45 min. | 55.6 | 16.8 | 36.2 |
| 60 min. | 58.6 | 13.2 | 35.9 |
| 75 min. | 55.0 | 11.9 | 33.5 |
| 90 min. | 54.8 | 17.6 | 36.2 |
| 90° - 100° C | | | |
| 15 min. | 54.5 | 14.7 | 34.6 |
| 30 min. | 50.6 | 14.8 | 32.7 |
| 45 min. | 50.8 | 15.8 | 33.3 |
| 60 min. | 53.9 | 13.0 | 35.5 |
| 75 min. | 50.3 | 16.4 | 33.4 |
| 90 min. | 51.2 | 14.1 | 32.7 |

L.S.D. for comparing pretreatment means within seed lots,
at 0.05 level = 4.8.
at 0.01 level = 6.4.

L.S.D. for comparing pretreatment means over both seed lots,
at 0.05 level = 3.4.
at 0.01 level = 4.5.

C.V. = 57.1%.

Table 8. Analysis of variance of germination following pre-treatment with hydrogen peroxide, and the high dry and low moist pretreatments.

| Source | D.F. | S.S. | M.S. | F |
|----------------------|------|----------|----------|-----------|
| Total | 103 | 45040.16 | 437.28 | |
| Treatment | 25 | 44270.13 | 1770.81 | 196.32** |
| Lots | (1) | 43614.81 | 43614.81 | 4835.34** |
| Pretreatments | (12) | 419.99 | 34.99 | 3.88** |
| Lots X Pretreatments | 3 | 235.33 | 19.61 | 2.17** |
| Blocks | 75 | 99.66 | 33.22 | 3.68** |
| Error | | 676.37 | 9.02 | |

** Significant at 0.01 level.

Table 9. Mean germination (arc sin $\sqrt{\%}$) following pretreatment with hydrogen peroxide, and the high dry and low moist pretreatments.

| Pretreatment | Within seed lots | | Over both seed lots |
|------------------|------------------|-------|---------------------|
| | Lot E | Lot H | |
| High dry | 53.4 | 14.8 | 34.1 |
| Low moist | 60.2 | 21.7 | 41.0 |
| Soaking in water | 52.7 | 14.2 | 33.5 |
| 1% H_2O_2 | | | |
| 1 min. | 60.1 | 13.1 | 36.6 |
| 2 min. | 53.2 | 14.6 | 33.9 |
| 4 min. | 58.6 | 13.6 | 36.1 |
| 8 min. | 58.6 | 14.6 | 36.6 |
| 15 min. | 52.9 | 15.0 | 35.0 |
| 30 min. | 56.1 | 17.9 | 37.0 |
| 60 min. | 56.7 | 12.1 | 34.4 |
| 5% H_2O_2 | | | |
| 15 min. | 57.1 | 17.5 | 37.5 |
| 30 min. | 57.3 | 15.5 | 36.4 |
| 60 min. | 52.7 | 14.0 | 34.2 |

L.S.D. for comparing pretreatment means within seed lots,
at 0.05 level = 4.2.
at 0.01 level = 5.6.

L.S.D. for comparing pretreatment means over both seed lots,
at 0.05 level = 3.0.
at 0.01 level = 4.0.

C.V. = 58.4%.

peroxide pretreatment greater than that following the low moist pretreatment.

Soaking in Aqueous Solution of Sodium Hypochlorite (Clorox)

Results are summarized in Tables 10 and 11. In the analysis of variance the term, times, is used to designate durations of sodium hypochlorite pretreatment. The high dry and low moist pretreatment, and soaking for 60 minutes in distilled water, are referred to as checks.

Highly significant differences occurred between seed lots, among the three check pretreatments, and between the checks and various durations of sodium hypochlorite pretreatment. The interaction, seed lots by checks, was significant at the 0.05 level.

In no case was germination significantly improved by the sodium hypochlorite pretreatment. The low moist pretreatment significantly improved germination in lot H but not in lot E.

Hull Removal

Results are summarized in Tables 12 and 13. Highly significant differences occurred between seed lots and among pretreatments. The interaction, seed lots by pretreatments, was also highly significant.

Table 10. Analysis of variance of germination following pretreatment with sodium hypochlorite, and the high dry and low moist pretreatments.

| Source | D.F. | S.S. | M.S. | F. |
|-----------------------------|------|----------|----------|----------|
| Total | 55 | 21756.27 | 395.57 | |
| Treatment | 13 | 20816.99 | 1601.31 | 63.61** |
| Seed Lots | 1 | 19273.45 | 19273.45 | 825.77** |
| Pretreatments | 6 | 1123.46 | 187.24 | 8.02** |
| Among Checks | (2) | 578.84 | 289.43 | 12.40** |
| Among Times | (3) | 102.76 | 34.25 | 1.47 |
| Checks vs. Times | (1) | 441.84 | 441.84 | 18.93** |
| Seed Lots X Pretreatments | 6 | 420.08 | 70.01 | 3.00* |
| Seed Lots X Checks | (2) | 186.66 | 93.33 | 3.99* |
| Seed Lots X Times | (3) | 226.44 | 9.70 | 0.42 |
| Seed Lots X Check vs. Times | (1) | 6.94 | 6.94 | 0.30 |
| Blocks | 3 | 29.13 | 9.71 | 0.42 |
| Error | 39 | 910.15 | 23.34 | |

* Significant at 0.05 level.

** Significant at 0.01 level.

Table 11. Mean germination (arc sin $\sqrt{\%}$) following pretreatment with sodium hypochlorite, and the high dry and low moist pretreatments.

| Pretreatment | Within seed lots | | Over both seed lots |
|-----------------------------|------------------|-------|---------------------|
| | Lot E | Lot H | |
| High dry | 55.4 | 12.8 | 34.1 |
| Distilled water | 57.5 | 16.4 | 37.0 |
| Low moist | 60.7 | 30.6 | 45.7 |
| Sodium hypochlorite 15 min. | 57.4 | 14.2 | 35.8 |
| 30 min. | 44.9 | 16.6 | 30.3 |
| 45 min. | 52.3 | 14.3 | 33.3 |
| 60 min. | 51.3 | 14.9 | 35.1 |

L.S.D. for comparing pretreatment means within seed lots,

at 0.05 level = 6.9.

at 0.01 level = 9.3.

L.S.D. for comparing pretreatment means over both seed lots,

at 0.05 level = 4.9.

at 0.01 level = 6.6.

C.V. = 55.6%

Table 12. Analysis of variance of germination following hull removal, and the high dry and low moist pretreatments.

| Source | D.F. | S.S. | M.S. | F |
|----------------------|------|----------|----------|-----------|
| Total | 25 | 14358.92 | 624.30 | |
| Treatment | 5 | 14199.51 | 2839.90 | 273.07** |
| Lots | (1) | 13575.53 | 13575.53 | 1305.34** |
| Pretreatments | (2) | 481.50 | 240.75 | 23.15** |
| Lots X Pretreatments | (2) | 142.48 | 71.24 | 6.85** |
| Blocks | 3 | 5.45 | 1.15 | 0.11 |
| Error | 15 | 155.96 | 10.40 | |

** Significant at 0.01 level.

Table 13. Mean germination (arc sin $\sqrt{\%}$) following hull removal, and the high dry and low moist pretreatments.

| Pretreatment | Within seed lots | | Over both seed lots |
|--------------|------------------|-------|---------------------|
| | Lot E | Lot G | |
| High dry | 52.6 | 0.6 | 26.6 |
| Low moist | 60.6 | 10.6 | 35.6 |
| Hull removal | 56.9 | 16.1 | 36.5 |

L.S.D. for comparing pretreatment means within seed lots,

at 0.05 level = 4.9.

at 0.01 level = 6.7.

L.S.D. for comparing pretreatment means over both seed lots,

at 0.05 level = 3.5.

at 0.01 level = 4.8.

C.V. = 76.4%.

Hull removal significantly improved germination in lot H but not in lot E. The low moist pretreatment significantly improved germination in both lots. In lot H, germination following hull removal was significantly higher than that following the low moist pretreatment.

Treatment with Gibberelllic Acid

Results are summarized in Tables 14 and 15. Highly significant differences occurred between seed lots and between the high dry and low moist checks. The interactions, seed lots by between checks and seed lots by among concentrations, were also highly significant.

Treatment with gibberelllic acid at concentrations of 50mg/liter, 75mg/liter, and 100mg/liter significantly increased germination in lot H. Germination of lot E was not significantly affected by treatment with gibberelllic acid, although a reduction in germination at a concentration of 100mg/liter approached significance at the 0.05 level. The low moist pretreatment significantly improved germination in lot H but not in lot E.

Relation of Caryopsis Moisture

Uptake to Seed Dormancy

Results are summarized in Table 16. Highly dormant seed lot H and less dormant seed lot E did not differ significantly in water uptake.

Table 14. Analysis of variance of germination in aqueous solution of gibberellic acid, and following the high dry and low moist pretreatments.

| Source | D.F. | S.S. | M.S. | F |
|--|------|----------|----------|-----------|
| Total | 47 | 15478.76 | 329.33 | 116.02** |
| Treatments | 11 | 15054.87 | 1368.62 | 1229.07** |
| Seed Lots | 1 | 14490.75 | 14490.75 | 3.54* |
| Pretreatments and G.A. | | | | |
| Treatment | 5 | 208.55 | 41.71 | 14.17** |
| Between Checks | (1) | 167.06 | 167.06 | 1.17 |
| Among Concentrations | (3) | 41.45 | 13.82 | 0.003 |
| Check vs. Concentrations | (1) | 0.04 | 0.04 | 6.03** |
| Seed Lots X Pretreatments | 5 | 355.57 | 71.11 | 15.20** |
| Seed Lots X Between Checks | (1) | 185.64 | 185.64 | 4.76** |
| Seed Lots X Among Concentra- tions | (3) | 168.20 | 56.07 | 0.15 |
| Seed Lots X Checks vs. Concentrations | (1) | 1.73 | 1.73 | 0.98 |
| Blocks | 3 | 34.63 | 11.54 | |
| Error | 33 | 389.26 | 11.79 | |

* Significant at 0.05 level.

** Significant at 0.01 level.

Table 15. Mean germination (arc sin $\sqrt{\%}$) in aqueous solution of gibberellic acid, and the high dry and low moist pretreatments.

| Pretreatment and Treatment | Within seed lots | | Over both seed lots |
|----------------------------|------------------|-------|---------------------|
| | Lot E | Lot H | |
| High dry | 57.3 | 15.2 | 36.3 |
| Low moist | 56.9 | 28.5 | 42.7 |
| Gibberellic acid | | | |
| 25 mg/l | 59.9 | 18.2 | 39.1 |
| 50 mg/l | 56.5 | 24.3 | 40.4 |
| 75 mg/l | 58.3 | 23.5 | 40.9 |
| 100 mg/l | 52.6 | 23.4 | 38.0 |

L.S.D. for comparing means within seed lots,

at 0.05 level = 4.9.

at 0.01 level = 6.7.

L.S.D. for comparing means over both seed lots,

at 0.05 level = 3.5.

at 0.01 level = 4.8.

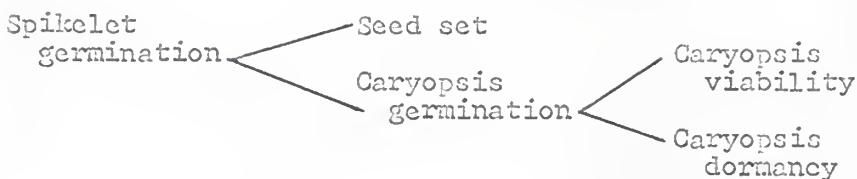
C.V. = 45.8%.

Table 16. Water uptake of samples of 100 cryopcs.

| | Seed lot Mean | | Mean difference | u |
|--|---------------|-------|-----------------|------|
| | E | H | | |
| Initial weight (g) | 0.224 | 0.141 | 0.083 | |
| Wet weight (g) | 0.290 | 0.194 | 0.096 | |
| % water absorbed, based on initial weight | 29.5 | 37.6 | 8.1 | 0.25 |
| Oven dry weight (g) | 0.220 | 0.136 | 0.084 | |
| % water absorbed, based on oven-dry weight | 31.8 | 42.6 | 10.08 | 0.31 |

DISCUSSION AND CONCLUSIONS

Spikelet germination is a composite trait involving the effects of seed set, caryopsis viability, and caryopsis dormancy. The interrelationships of these traits may be represented schematically as follows:



In this study, random variation in seed set may have obscured real treatment effects by contributing to error in the measurement of spikelet germination. In six of the seven experiments the coefficient of variability was greater than 40 percent, being 76.4 percent in the experiment on hull removal. Elimination of the influence of seed set through use of samples of free caryopses would probably have reduced the coefficient of variability, but, since hull removal has been shown to affect germination, such procedure would have been unadvisable. Measurement of the caryopsis content of individual germination samples, and subsequent removal of the influence of seed set through covariance analysis, would probably have increased the precision of the experiments. The caryopsis content of each sample could have been determined prior to germination by means of an x-ray technique recently described by Barnett and Rafii¹. Hull removal following

¹Barnett, F. L., Personal communication.

germination would also have permitted an estimation of seed set as the sum of the numbers of germinated and ungerminated caryopses. Hull removal is very time consuming, however, and would not have been feasible in this study. The influence of random variation in seed set could have been reduced through use of larger samples or through increased replication.

Some of the pretreatments, notably the high-immersion and high-temperature procedures, obviously result in some loss in caryopsis viability. The effect of any pretreatment can, in fact, be considered to consist of a viability-reducing component and a dormancy-reducing component, the net effect of the pretreatment depending upon the relative magnitudes of the two component effects. Thus, failure to obtain increased germination following a given pretreatment should be interpreted as indicating only that the dormancy-reducing component is not greater than the viability-reducing component--not that the former component is zero.

Consideration of these relationships is important in interpretation of the high-temperature and hull-removal results. Temperatures of 90-100°C for 30, 45, 75, and 90 minutes had a viability-reducing effect as evidenced by the significant reduction in germination in lot E. Since germination in the presumably more dormant lot H was not reduced, it seems likely that, in this lot, reduction in viability was offset by a reduction in dormancy--a reduction not possible in the less dormant lot E. Hence, it would appear that high temperature has a dormancy-reducing effect

but that this effect is generally obscured by a reduction in viability. The possibility that lot H was simply less susceptible to high temperature than was lot E must also be considered, however. Hull removal has a pronounced dormancy-reducing effect as evidenced by the fact that this pretreatment was more effective than the low moist pretreatment in increasing germination in lot H. That it compared less favorably with the low moist pretreatment in lot E, however, suggests that hull removal has a viability-reducing effect which may obscure its dormancy-reducing effect at low levels of dormancy. Evaluation of a pretreatment with respect to its dormancy-reducing effect only would obviously be difficult and would generally require some method other than germination for determination of caryopsis viability.

Of all pretreatments evaluated, exposure to low temperature in a moistened condition and hull removal are most promising. Practical application of these pretreatments, on anything but a laboratory scale can, of course, be questioned. In this connection, however, it should be noted that a relatively complicated dormancy-breaking treatment, involving soaking in aqueous solution of potassium nitrate and subsequent exposure to low temperature, is commonly used on a commercial basis with seed of buffalograss, Buchloe dactyloides (Nutt.) Engelm. (Wheeler and Hill, 1957). Further, should the effectiveness of hull removal be found due to caryopsis scarification, other methods of scarification, such as those involving use of acids, might be more practical.

Results of this study do not provide a sound basis for the

development of a theory concerning the mechanism of seed dormancy in indiangrass. They are of interest, however, in comparison with results of other workers and in connection with theories of dormancy already proposed for indiangrass and other species. Coukos (1944) stated that seed coats of some native grasses (including indiangrass) permit entrance of water even in unscarified seeds. He suggested that the cause of dormancy might involve gas-exchange restrictions in the seed coats. Meyer et al. (1960) suggested that effectiveness of low temperature treatment is associated in some species with a favorable relation between respiration rate and rate of oxygen absorption or carbon dioxide liberation. However, Koller et al. (1962) stated that the mechanism of low temperature is obscure. Effectiveness of hull removal in this study suggests involvement of permeability factors, since removals of chaffy appendages almost invariably results in scarification of the pericarp and probably also the testa, and would presumably increase the permeability of these coverings. Shimizu (1959) reported that dormant seeds of erabgrass, Digitaria adscendens (HBK) Henr., absorbed little water. He found a period of after-ripening was necessary to permit entry of water in sufficient quantity to support germination. Since, in this study, caryopses of the less dormant seed lot E and the highly dormant seed lot H did not differ in water uptake, however, it would seem that water permeability of the pericarp and testa is not involved. Results of the experiment on water uptake, on the other hand, do not preclude involvement of permeability to such gases as oxygen and carbon

dioxide, nor do they preclude involvement of water permeability of the chaffy appendages which were removed at the beginning of the experiment. Involvement of permeability to oxygen and carbon dioxide may explain similarity of the effects of the low moist and hull removal pretreatments. The low moist procedure may reduce the need for gas exchange by reducing the rate of respiration while hull removal may increase the rate of gas exchange. According to Meyer et al. (1960), the low moist pretreatment may also induce permeability changes in the seed coats. It seems unlikely that the chaffy appendages of dormant spikelets are impermeable to either water or gases, although this possibility does not appear to have been investigated. Results of Barnett and Rafii¹ indicate that caryopsis dormancy is not always entirely overcome by either hull removal or low temperature pretreatment.

Westra and Loomis (1966) improved seed germination of Uniola paniculata L. by cutting deeply into the endosperm and subsequently leaching the seeds. They suggested that cutting and leaching released a diffusible inhibitor. A similar germination inhibitor might be involved in seed dormancy in indian-grass. During the low moist pretreatment such an inhibitor could be leached out of the spikelet permitting germination when temperature became favorable. Increased germination following hull removal could be due to removal of an inhibitor in the chaffy appendages or due to leaching of an inhibitor

¹ Barnett, F. L., Personal Communication.

from the caryopsis. According to Naylor and Simpson (1961), natural inhibition of germination in wild oats was overcome by treatment with gibberellic acid. In this study, gibberellic acid had some effect on the highly dormant seed lot H but failed to increase germination in lot E.

Demowsay (1916) improved germination of garden cress through treatment with hydrogen peroxide. He proposed that the peroxide had an oxidation effect upon the seed coat. Sumner and Cobb (1962) reported that sodium hypochlorite was effective in removing a germination inhibitor in seeds of Coronado sideoats grama. Both hydrogen peroxide and sodium hypochlorite are oxidizing agents. Their effect on seeds of indiangrass was unimpressive, hydrogen peroxide having some effect on the less dormant lot E and sodium hypochlorite failing to produce a germination increase with either lot E or lot H.

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EVALUATION OF METHODS OF REDUCING SEED DORMANCY
IN INDIANGRASS, SORGHASTRUM NUTANS (L.) NASH

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Spikelets of indiangrass, (Sorghastrum nutans (L.) Nash), of varying age and background were subjected to a number of germination and pregermination procedures in an attempt to evaluate the effectiveness of various treatments in alleviating seed dormancy. Pregermination treatments included: soaking in water at room temperature and at 5°C, dry storage at room temperature and at 5°C, storage in a moistened condition at 5°C, exposure to high temperature, soaking in aqueous solution of hydrogen peroxide, soaking in aqueous solution of sodium hypochlorite, and hull removal. A single germination treatment involved application of gibberellic acid.

Germination was generally improved following storage for two weeks in a moistened condition at 5°C. With highly dormant seed, hull removal was more effective than the foregoing pretreatment; with less dormant seed, however, results following the two pretreatments were not significantly different. Germination increase following the use of hydrogen peroxide and gibberellic acid was unimpressive. Other procedures resulted in no significant increase in germination.

Random variation in seed set may have obscured real treatment effects by contributing to error in the measurement of spikelet germination. Some of the pretreatments, notably soaking in water at room temperature and exposure to high temperature, obviously resulted in some loss in caryopsis viability. The effect of any pretreatment can be considered to consist of a viability-reducing component and a dormancy-reducing component;

the net effect of the pretreatment depending upon the relative magnitude of the two component effects. In some instances where a given pretreatment or treatment appeared ineffective, real dormancy-reducing effects may have been obscured by loss in caryopsis viability.

Similarity of the effects of hull removal and exposure to low temperature in a moist condition may indicate involvement of seed covering permeability to gas exchange. Exposure to low temperature in a moist condition may reduce the need for gas exchange by reducing the rate of respiration, while hull removal may increase the rate of gas exchange. Leaching of a germination inhibitor may also be involved. Since highly dormant and less dormant caryopses did not differ in water uptake, it seems unlikely that water permeability of either the pericarp or the testa is involved in seed dormancy of indiangrass.